POLYSACCHARIDE-BASED 3D PRINTING INKS SUPPLEMENTED WITH ADDITIVES

Alexandra CERNENCU[1], Adriana LUNGU[2], Izabela Cristina STANCU[3], Eugeniu VASILE[4], Horia IOVU[5]

The design of 3D printable formulations by combining different polysaccharides with effective additives is the key direction of the current paper. Promising bi-component inks were formulated using one type of cellulose (carboxymethylcellulose or nanocellulose) in combination with sodium alginate, a natural polysaccharide isolated from marine algae. To attain a good printability a proper selection of the ink components was carried out. Moreover, suitable additives were employed to modulate specific physical properties. For the preparation of the printable inks a wetting agent (glycerol / PEG) was added. Therefore, this work provides a comprehensive study of the additives' impact over the rheological and printing properties of polysaccharides-based-materials.

Keywords: polysaccharides based-inks, 3D printing, wetting agents

1. Introduction

Today, three-dimensional (3D) design technology is progressively broadening its applications in various fields of industry. The printability of a material is mainly determined by the printing technique and a material is defined as printable when the spatial deposition is well controlled and the object preserves the designed shape [1]. The printing process is challenged by various rheological and chemical features of the material. A wide range of materials have shown potential in 3D printing but modeling these materials into self-supporting items with tunable mechanical properties, degradation, and suitable rheological response is still an issue.

[1] PhD student, Department of Bioresources and Polymer Science, Faculty of Applied Chemistry and Materials Science, Advanced Polymer Materials Group, University "POLITEHNICA" of Bucharest, Romania, e-mail: alex.cernencu@gmail.com

[2] Prof., Department of Bioresources and Polymer Science, Faculty of Applied Chemistry and Materials Science, Advanced Polymer Materials Group, and Faculty of Medical Engineering, University "POLITEHNICA" of Bucharest, Romania, e-mail: adriana_lungu2006@yahoo.com

[3] Prof., Department of Bioresources and Polymer Science, Faculty of Applied Chemistry and Materials Science, Advanced Polymer Materials Group, and and Faculty of Medical Engineering University "POLITEHNICA" of Bucharest, Romania, e-mail: izabela.cristina.stancu@gmail.com

[4] Prof. Department of Science and Engineering of Oxide Materials and Nanomaterials, University Politehnica of Bucharest, Romania, e-mail: eugeniuvasile@yahoo.com

[5] Prof. Department of Bioresources and Polymer Science, Faculty of Applied Chemistry and Materials Science, Advanced Polymer Materials Group, University "POLITEHNICA" of Bucharest, Romania, e-mail: horia.iovu@upb.ro

Natural-derived polymers are an inexhaustible source of inspiration in the development of printable materials as they offer distinct advantages in terms of chemical versatility and flowing properties that govern the printing process. Among them, polysaccharides (e.g. alginate, cellulose, dextran etc.) have been investigated for their potential as 3D printable inks [2]. Sodium alginate (Alg) is widely used in 3D bio-printing within the last years due to its outstanding ability to achieve an excellent gelation in the presence of divalent ions which helps to better preserve the shape after extrusion [3]. However, pristine solutions of Alg exhibit poor printability so that the use of rheological modifiers which induce increased viscosity and modulate the shear-thinning behavior is prevalent in order to improve both shape fidelity and stability. In the recent years, several papers described the additive manufacturing of scaffolds with accurate geometries using hybrid hydrogels of alginate combined with various components especially of polymeric nature [4, 5].

As a rheological modifier, plant-derived cellulose stands out as a valuable compound, drawing an increasing attention towards the study of its thickening and rheological properties [6]. One of the water-soluble versions of cellulose is carboxymethylcellulose (CMC), well-known for its uses as viscosity modifier [7] for various applications including 3D prints. Notwithstanding, cellulose nanofibers (CN) hold a great potential in 3D printing as they provide a pronounced shear-thinning behavior to the printing material, this rheological phenomenon enhancing the printability and the shape fidelity after extrusion.

A lot of additional components like wetting agents, dispersing agents, antifoaming agents etc. can be added to printing formulations to prepare performant inks adapted to the required printing technology. Careful selection of suitable additives would represent a useful approach to deal with essential problems associated with printable water-based polymeric inks mainly the difficulties to modulate rheological and wetting properties. Moreover, formulations additives must be water-soluble in order to formulate high-performance inks envisaged for tissue engineering. The current research paper is focused on the study of incorporation effect of a wetting agent (also known as humectant), within ink prior being dispensed by the 3D printer. In general, these particular additives improve the wettability of the ink, reduce the contact angle that occurs between (bio)ink and the printer syringe/nozzle [8] and also prevent the ink composition from drying out or crusting in the nozzle. Representative examples of such humectants which may be used in aqueous-based ink compositions include: polyethylene glycol (PEG), glycerol (Gly), sorbitol, urea etc.[9]. Although Gly and PEG are widely used as wetting agents for biopolymers, within cellulose derivatives as well [10], their significance in 3D (bio)printing field was underexploited and much less in ink formulations based on Alg combined with cellulosic materials. To the best of our knowledge, Gly was only

assessed in CN/Alg printing formulations [11] while CMC/Alg blends were just recently reported as suitable in 3D printing [12]. Yet the effect of PEG addition in such ink formulations has not been studied and a comparative research is envisaged to encourage the design of new performant ink formulations.

The current paper describes promising ink formulations based on CMC/Alg and CN/Alg, bringing in the foreground the contribution of additives. A small organic molecule (Gly) and a low-molecular weight polymer (PEG) were used as wetting agents for the preparation of an array of bi-component inks. Through a comparative study of CMC and CN as precursor inks, the influence of additives over the rheological and printing properties of precursor inks was assessed.

2. Experimental data

2.1. Materials

Sodium alginate (alginic acid sodium salt from brown algae with a mannuronic acid to guluronic acid (MG) ratio of 1.5) and sodium carboxymethyl cellulose (average Mw ~90,000) were purchased from Sigma-Aldrich. Never-dried bleached sulfite pulp from hardwood with a 13% solid content was kindly provided by Stora Enso, Sweden and used for the synthesis of CN as component of printable inks. All the chemicals and reagents used in the process of producing CN: 2,2,6,6-tetramethylpiperidine 1-oxyl (TEMPO) sodium bromide (NaBr), sodium hydroxide (NaOH) 98% (pellets), hydrochloric acid (HCl) ACS reagent, 37%, sodium hypochlorite solution (NaClO) (6-14% active chlorine – supplier description; 12% active chlorine – experimental value by iodometric determination) EMPLURA® and calcium chloride ($CaCl_2$) were purchased from Sigma-Aldrich.

Low-molecular weight compounds, glycerol (M_w 92.09 g/mol) and polyethylene glycol (PEG) (M_w 200 g/mol) were supplied from Sigma-Aldrich and used to supplement the precursor inks.

2.2. Methods

Synthesis of CN suspension. The CN suspension has been produced using the protocol described elsewhere [13]. Briefly, the never-dried cellulose pulp (1 g of cellulose solid content) was suspended in water (100 mL) containing TEMPO (0.0125 g) and NaBr (0.125 g). NaClO (2.5 mmol) was then added and the oxidation reaction was conducted at room temperature under gentle agitation. The pH was maintained at 10.5 by adding 0.5 M NaOH for 1.5 h and then adjusted to 7 by adding 0.5 M HCl. The oxidized pulp was thoroughly washed and further subjected to mechanical disintegration by sonication treatment applied to the cellulose/water slurry. Thereby, through oxidative treatment, CN was produced as dense and transparent gel-like dispersion with a solid content of 1.09%.

Ink preparation. Printing inks were formulated by using two precursor mixtures, CMC:Alg and CN:Alg in 1:1 mass ratio. For the first mentioned formulations, a solution of 4% CMC was prepared in distilled water and the corresponding amount of Alg was added under gentle stirring. The latter formulations were obtained by directly dissolving Alg within CN suspension. Several ink formulations were further prepared by using two types of humectants (PEG and Gly) in 5% concentration and the composition of each formulation is described in Table 1.

Table 1.

Composition of ink formulations

Composition	Gly (%)	PEG (%)	Sample code
CMC:Alg	-	-	CMC-A
CMC:Alg	-	5	CMC-A#PEG
CMC:Alg	5	-	CMC-A#Gly
CN:Alg	-	-	CN-A
CN:Alg	-	5	CN-A#PEG
CN:Alg	5	-	CN-A#Gly

2.3. Equipments

Fourier Transform Infrared Spectroscopy (FTIR) measurements were performed on a Bruker Vertex 70 FTIR spectrometer equipped with an attenuated total reflectance (ATR) accessory. Laboratory conductivitymeter (WTW inoLab® Cond 7110) was used for the determination of carboxyl content using the conductometric titration method according to SCAN 65:02. For Transmission Electron Microscopy (TEM) observations, drops of dilute CN suspension (0.1% solid content) were deposited on to carbon-coated electron microscopy grids and images were collected by means of a Tecnai^{TM} G2 F30 S-TWIN transmission electron microscope. The steady shear viscosity of the aqueous solutions of the precursors was measured on a Kinexus Pro rheometer (Malvern Instruments) equipped with a Peltier element for temperature control. The shear viscosity of the precursors suspensions was measured using the cone-plate geometry (cone angle of 4^{o} and a diameter 40 mm) at 25^{o}C increasing the shear rate from 0.01 to 1000 s^{-1}.

The ink formulations were printed using the 3D bioprinter 3D Discovery^{TM} from RegenHU (Switzerland). The direct dispensing print head was used to conduct the preliminary printing experiments as well as 3D printing of hydrogel scaffolds. The ink formulations were printed using a G25 needle with a 250 μm inner diameter. In order to determine the influence of the printing pressure and feed rate over the printed line width, for each formulation, lines having 20 mm length were deposited with varied feed rate (from 5 to 40 mm/s, with 5 units' step). For the 3D constructs the G-codes were generated by the use of the

BioCAD^{TM} software, as square grids of 20 mm x 20 mm with a crosshatch infill of 3 mm spacing between lines in 10 layers with a 0^o-90^o deposition direction. The 3D constructs were printed directly onto glass slides at room temperature (25^{o}C).

3. Results and discussion

Alg as pure material exhibits poor applicability in 3D direct writing and so forth CMC and CN were employed to enhance the viscosity; moreover, due to the presence of -COOH groups, the cellulosic component together with Alg engage in the ionic crosslinking process envisaged for the printed grids. The cellulose -OH groups modification has a tremendous effect over its properties especially in terms of solubility, rheological and mechanical behavior. Both cellulose types used herein, CMC and CN, differ greatly with respect to chemical and morphological features and yet both present favorable printing behavior.

3.1. Determination of structural characteristics of CN

To obtain printable CN, a highly selective oxidation of cellulose pulp was performed according to Section 2 and fully characterized to study their structural features as described further on. A schematic draw illustrating the organization of cellulosic fibers before and after TEMPO-mediated oxidation is shown in Fig 1. The electrostatic repulsions and osmotic effects between the ionically charged carboxyl groups present on CN surfaces and water elements cause an individualized organization of the fibers, leading to the formation of a highly viscous material [14]. As depicted in Fig. 1, the outcome of oxidation reaction is a transparent gel with promising printing properties.

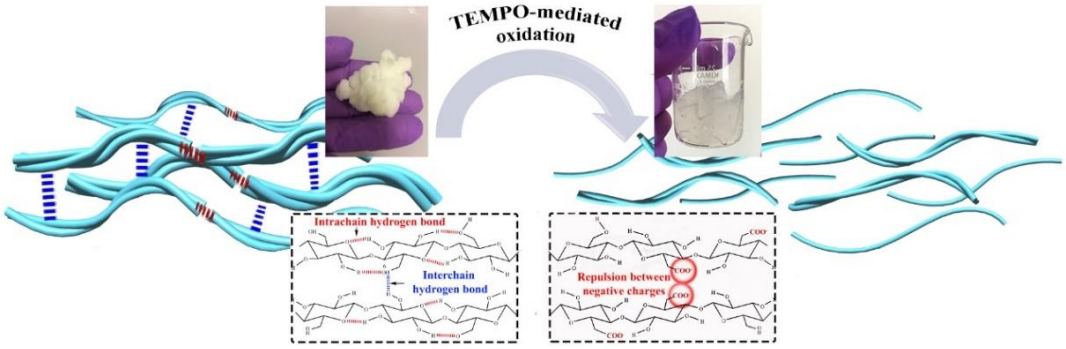

Fig. 1. Schematic representation of the fibrillary organization of cellulose before and after oxidation process

a) The oxidized CN were firstly evaluated by FT-IR spectrometry, to emphasize the structural changes with respect to the cellulose native state (Fig 2.).

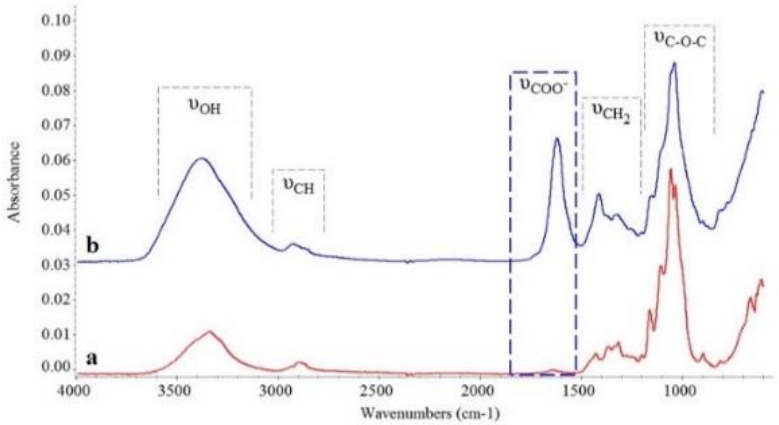

Fig. 2. The FTIR spectra of never-dried bleached cellulose pulp (a) and CN (b)

Overall, both FTIR spectra revealed the OH stretching vibration band at around 3340 cm^{-1}, the C–H stretching signals around 2900 cm^{-1} and in the range 1420–1320 cm^{-1} signals that are associated with the deformation and stretching vibrations of CH_2 and the deformation vibration of the C-H groups. The strong and sharp peak around 1050 cm^{-1} is assigned to the stretching vibrations of the pyranose ring skeleton. Significant differences in the FTIR spectra of cellulose before and after oxidation treatment can be observed in the 1500-1800 cm^{-1} domain. The CN spectrum displays a sharp band at 1619 cm^{-1} assigned to C=O stretching vibration from $-COO^-Na^+$ groups [14]. The carboxylic group occurrence was thus qualitatively proved by FT-IR.

b) The surface chemistry of CN was further investigated in terms of oxidation extent of the -OH groups. The conductometric titration was used to quantify the total amount of carboxyl groups in cellulose fibers. The titration curve was plotted and the total carboxyl content (*X*) was calculated applying eq. 1.

$$X(\mu mol/g) = \frac{c\,(\mu mol/L)\cdot V(L)}{m\,(g)} \quad (1)$$

The precise concentration of NaOH solution (*c*) was determined prior to titration, while the dry-weight (m) of the sample was measured post-titration. The volume of NaOH consumed for neutralization (*V*) of carboxylic groups was measured at the intersection of the curve tangents. The total acidic content (835 µmol/g) indicated a significant number of carboxylate groups on the CN surface as also asserted by previous studies [15].

c) The air-dried CN suspension was diluted to 0.1% solid content and was visualized through TEM (Fig 3.).

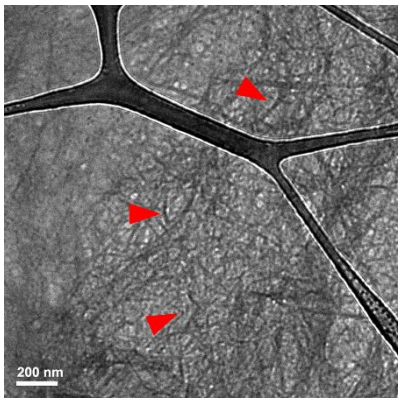

Fig. 3. TEM images of CN

The sample reveals highly entangled assemblies of individual fibers whose diameters were determined using ImageJ software in the range of 5 -10 nm. Neither the length nor length distribution of the nanofibrils can be accurately measured by TEM due to the dense fibrillar tangle formed and the high length/diameter ratio. Considering the final characteristics of thusly synthetized CN, it was further employed in the formulation of bi-component ink precursors.

3.2. Rheological evaluation of ink precursors

CMC and previously obtained CN were henceforth used in combination with Alg to develop polysaccharide-based formulations and the effect of the additives was investigated. Rheological measurements were employed to evaluate the viscosity of the precursor inks supplemented with a wetting agent. The relationship between the viscosity and shear rate is illustrated in Fig 4.

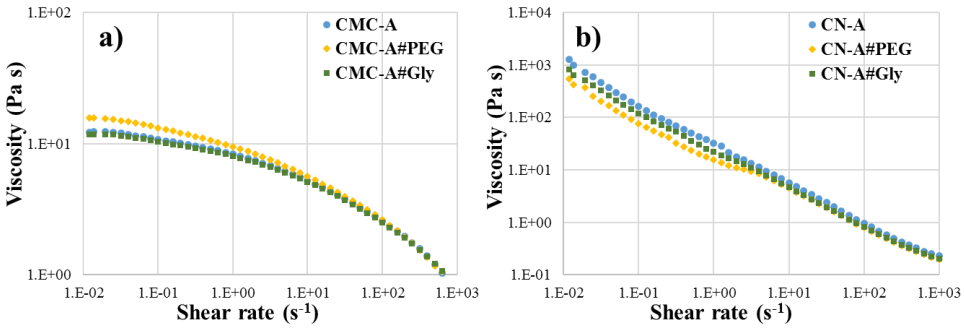

Fig. 4. Steady state-viscosity as a function of shear rates for CMC-A(a) and CN-A (b) formulations.

The shear viscosity of all blends shows a decreasing tendency as the shear rate increases, regardless the type of cellulose used. This rheological phenomenon, described as shear-thinning behavior, is of interest when assessing the flowing properties of formulations intended for 3D printing. In Fig 4a, the shear viscosity of the CMC formulations exhibits a slight decrease in the range of

0.01-1 s^{-1} while at higher shear rates, the shear thinning behavior is observed. The polymer chains exhibit the tendency to orientate in the flow direction when the shear rate is increased, so that the material meets less resistance to flow. Even though CMC-A formulations show shear-thinning behavior, there is no evidence of a yield stress, as a consequence of CMC input [16]. On the other hand, viscosity curves of CN precursors (Fig 4b) show shear-thinning behavior with a sudden drop in viscosity against increasing shear rate. The CN formulations display an evident power-law behavior within the measured shear rate range in good agreement with CN rheological response described in other research works [17] so that all formulations exhibit yield flow behavior. When investigating the effect of additives, it can be observed that for CMC compositions, PEG and Gly exhibit an opposite effect: the viscosity of CMC-A#PEG is increased while CMC-A#Gly shows a slightly lower viscosity related to CMC-A reference precursor. In parallel for CN composition, both PEG and Gly decrease the viscosity. It is well-known that this class of additives tends to decrease the ink viscosity while the shear-thinning properties are preserved [11, 18].

3.3. Experimental studies on the printing parameters for an optimum set-up.

Several printing tests were conducted to evaluate the inks printability and to define the optimum parameters that influence the printing resolution of 3D constructs. Therefore, a simple design of fully parametric parallel lines was used to evaluate the effect of printing pressure and speed on the line width. The printing tests were performed at 5 different pneumatic pressures from 100 to 140 kPa for CMC-based formulations and 40 to 80 kPa for CN-based formulations. Moreover, the printing speed was varied in the range of 5 - 40 mm/s for each pressure value. The widths of the extruded lines were considered only for continuous deposited filaments and are showed for comparison in Fig 5.

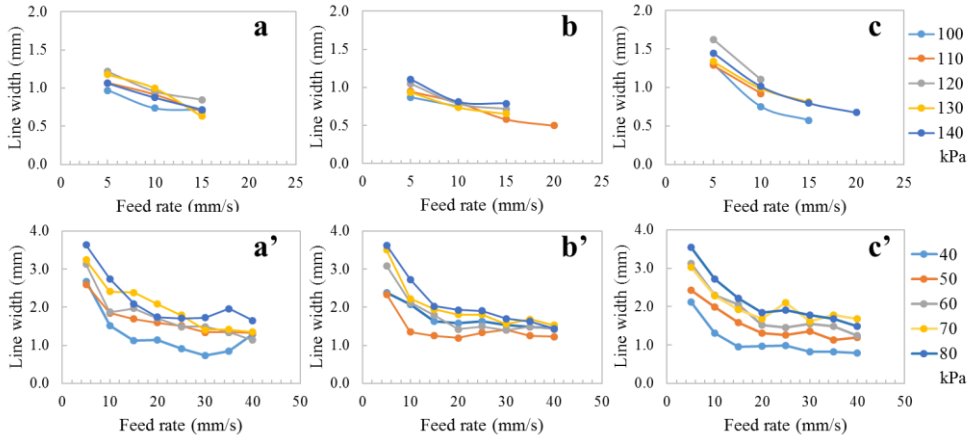

Fig. 5. Line widths of printed filaments under different combination sequences of printing parameters using CMC-A (a), CMC-A#PEG (b), CMC-A#Gly (c) and CN-A (a'), CN-A#PEG (b'), CN-A#Gly (c').

As the printing process is governed by the flow characteristics of the material, and consequently by its viscosity, different printing pressures were required for the extrusion of the studied formulations. It was found that CMC-A composition requires pressures over 100 kPa, while CN-A ink can be extruded at 40 kPa in spite of its higher viscosity. Considering their rheological profile, this can be explained by the discrepancy between their viscosity drop. It can be observed that by increasing the pressure flow, thicker lines are obtained regardless the precursor composition. On the other hand, the printing speed influences the tension along deposition direction so that at high feed rates the filaments become thinner. By comparing CMC and CN formulations, the former blends show lower filaments widths at all pressures. Yet, CMC formulations exhibit a narrow printing region as an increased pressure causes jetting-like extrusion which is unstable and hard to manage, while a too high printing speed leads to deposition of droplets instead of lines. Considering the best extrusion conditions, it can be observed that the printing speed and pressure exhibit an inversely proportional effect over the line thickness as the lowest diameters are recorded at low pressures and high printing speed. If CMC blends were printed at 10-15 mm/s the measured thickness of the printing lines was about 1 mm. By comparison, CN formulations show at the same printing speed line widths in the range of 1-2.5 mm depending on the pressure. Assessing the effect of additives, it can be observed that both improve the wetting properties leading to slightly lower filament diameters relative to the corresponding reference composition. Taking into consideration the optimum line width for each composition the process parameters were chosen to define the best set-up for the fabrication of 3D constructs (for CMC formulations: printing pressure 130±10 kPa at a feed-rate 15 mm/s; for CN formulations: printing pressure 50±5 kPa and feed-rate 20 mm/s)

3.4. Fabrication of polysaccharide-based scaffolds through 3D printing

3D printability of the studied formulation was assessed by choosing the optimum printing parameters to sequentially build lattice scaffolds. Pictures of the samples taken right after the printing process are shown in Fig 6. and their stability was studied in terms of strands resolution and pore geometries. Different water-soluble dyes were employed within the inks precursors to aid the visualization and indicate the differences between the formulations.

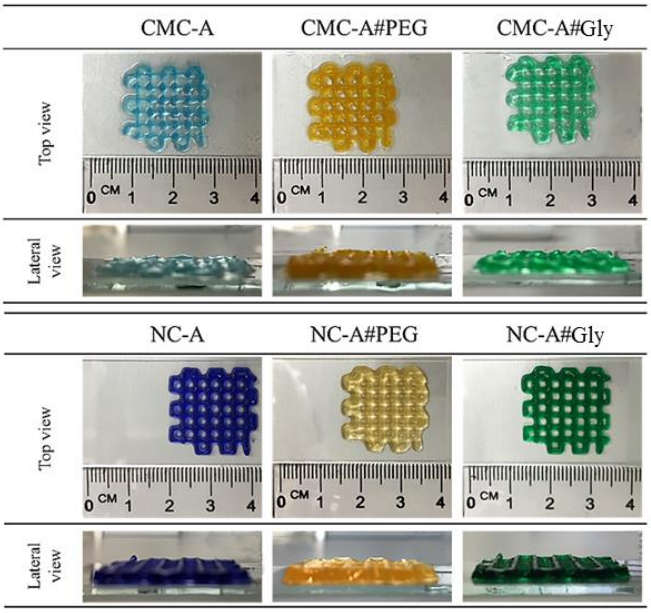

Fig. 6. Top-down view and lateral view of 3D printed objects with CMC-A, CMC-A#PEG, CMC-A#Gly and CN-A, CN-A#PEG, CN-A#Gly ink formulations

As confirmed by the measurements of the printed line widths, all the compositions could be translated to 3D scaffolds when optimum pressure and feed rate are applied. The grid structure of each sample can be clearly distinguished, and yet different printing resolutions can be observed. The scaffolds printed using CMC-A blends exhibit poor shape fidelity, showing material accumulation at strands intersection. The addition of PEG slightly improves the resolution and the printed strands become observable. However, it shows almost closed porosity due to diffusion caused by the gravity. A higher printing resolution is noticed for CMC-A#Gly, where the lattice is well distinguished. At the same time the area of crosshatch infill is irregular, and the strands show "rounded corners". In comparison with CMC formulations, CN inks exhibit superior printing properties through a better shape retention ability. CN-A reference sample shows a good strands distribution with almost circular pore geometry. Relative to the CN-A 3D construct, the additives exhibit opposing effects over printing resolution. When PEG was used, the material was poorly printed due to the strands fusion. The low viscosity affects the structural integrity of the filaments causing material overlapping. In contrast, when Gly was used, a better printing resolution was noticed. Qualitatively, the pore geometry is almost square and renders the path of the predesigned shape.

4. Conclusions

The cellulosic gels offer superior properties in the context of 3D direct writing, mainly in terms of rheological and mechanical behavior. Printable inks based on two distinct types of cellulose in combination with alginate were developed. The tested formulations help the fast formation of gel during fabrication and mixing cellulose with alginate will increase the solution viscosity which will improve the printability. The rheological evaluation showed that CMC compositions exhibit low viscosity and therefore they flow instantaneously upon application of stress, forming rather droplets than a continuous filament. The high viscosity drops registered for CN precursors facilitate the printing process as the resulting gel-like solution requires adequate external forces for formation of filament by direct extrusion though a nozzle. The extrusion rate depends on both printing pressure and nozzle feed rate and therefore parametric studies were conducted in order to evaluate the printing behavior of the formulations and the additives influence. Loading an additional compound (such as PEG and glycerol) suggested the improvement of the wetting properties leading to slightly lower filament diameters relative to the corresponding reference composition. Considering the printing performance with respect to the predesigned architecture it can be concluded that a successful 3D printing was achieved using glycerol in the ink composition. Therefore, the fabrication of 3D scaffolds using a cellulosic material combined with alginate and a suitable additive provide accurate geometries but also guide the applications of the obtained hybrid hydrogels in the biomedical field.

Acknowledgement

The 3D printing experiments were possible due to European Regional Development Fund through Competitiveness Operational Program 2014-2020, Priority axis 1, ID P_36_611, MySMIS code 107066, INOVABIOMED. A. Lungu would like to thank for the financial support provided by a grant of the Romanian Ministery of Research and Innovation, CCCDI – UEFISCDI, project number PN-III-P1-1.2-PCCDI-2017-0782 /REGMED – project 4 TUMOR, within PNCDI III. The work has been funded by the Operational Programme Human Capital of the Ministry of European Funds through the Financial Agreement 51668/09.07.2019, SMIS code 124705.

R E F E R E N C E S

[1]. *U. Jammalamadaka, K. Tappa*, "Recent advances in biomaterials for 3D printing and tissue engineering". in J. Funct. Biomater, **vol. 9**, no. 1, Mar. 2018, pp 22.

[2]. *F. Khan, S. R. Ahmad*, "Polysaccharides and their derivatives for versatile tissue engineering application". in Macromolecular Bioscience, **vol. 13**, no. 4, 2013, pp 395-421.

[3]. *D. Saha, S. Bhattacharya*, "Hydrocolloids as thickening and gelling agents in food: a critical review". in Journal of food science and technology, **vol. 47**, no. 6, 2010, pp 587-597.
[4]. *S. Sultan, G. Siqueira, T. Zimmermann, A. P. Mathew*, "3D printing of nano-cellulosic biomaterials for medical applications". in Current Opinion in Biomedical Engineering, **vol. 2**, no. 2017, pp 29-34.
[5]. *Q. Wang, J. Sun, Q. Yao, C. Ji, J. Liu, Q. Zhu*, "3D printing with cellulose materials". in Cellulose, **vol. 25**, no. 8, August. 2018, pp 4275-4301.
[6]. *C. Clasen, W.-M. Kulicke*, "Determination of viscoelastic and rheo-optical material functions of water-soluble cellulose derivatives". in Progress in Polymer Science, **vol. 26**, no. 9, 2001, pp 1839-1919.
[7]. *B.D. Braun, M. R. Rosen*, "Part 2 - Commercially available rheology modifiers", Rheology Modifiers Handbook, William Andrew Publishing , 1999, pp 71-191.
[8]. *D. Obradović, D. Obradović*, "Impact wetting and contact angle on quality offset press". in International Journal of Multidisciplinary Research and Development, **vol. 2**, no. 4, March. 2015, pp 27-31.
[9]. *A. A. Konta, M. García-Piña, D. R. Serrano*, "Personalised 3D printed medicines: which techniques andpolymers are more successful?". in Bioengineering, **vol. 4**, no. 4, 2017, pp 79.
[10]. *M. G. A Vieira, M. A. da Silva, L. O. dos Santos, M. M. Beppu*. "Natural-based plasticizers and biopolymer films: A review". European Polymer Journal, **vol 47**, no 3, 2011, pp 254-263.
[11]. *J. Leppiniemi, P. Lahtinen, A. Paajanen, R. Mahlberg, S. Metsä-Kortelainen, T. Pinomaa, H. Pajari, I. Vikholm-Lundin, P. Pursula, V. P. Hytönen*, "3D-printable bioactivated nanocellulose–alginate hydrogels". in ACS Applied Materials & Interfaces, **vol. 9**, no. 26, 2017, pp 21959-21970
[12]. A. Habib, V.Sathish, , S. Mallik, B. Khoda. "3D printability of alginate-carboxymethyl cellulose hydrogel". Materials, **vol. 11**, no. 3, 2018, pp 454-476.
[13]. *T. Saito, Y. Nishiyama, J.-L. Putaux, M. Vignon, A. Isogai*, "Homogeneous suspensions of individualized microfibrils from TEMPO-catalyzed oxidation of native cellulose". in Biomacromolecules, **vol. 7,** no. 6, 2006, pp 1687-1691.
[14]. *M. Shimizu, T. Saito, A. Isogai*, "Water-resistant and high oxygen-barrier nanocellulose films with interfibrillar cross-linkages formed through multivalent metal ions". in Journal of Membrane Science, **vol. 500**, no. 2016, pp 1-7.
[15]. *A. Rees, L. C. Powell, G. Chinga-Carrasco, D. T. Gethin, K. Syverud, K. E. Hill, D. W. Thomas*, "3D bioprinting of carboxymethylated-periodate oxidized nanocellulose constructs for wound dressing applications". in BioMed Research International, **vol. 2015**, no. 2015, pp.1-7
[16]. *A. Benchabane, K. Bekkour*, "Rheological properties of carboxymethyl cellulose (CMC) solutions". in Colloid and Polymer Science, **vol. 286**, no. 10, 2008, pp 1173.
[17]. *T. Moberg, K. Sahlin, K. Yao, S. Geng, G. Westman, Q. Zhou, K. Oksman, M. Rigdahl*, "Rheological properties of nanocellulose suspensions: effects of fibril/particle dimensions and surface characteristics". in Cellulose, **vol. 24**, no. 6, 2017, pp 2499-2510..
[18]. *A. P. Verrall, S. E. Brown.* "Carboxymethyl cellulose-based films and edible food casings made therefrom." U.S. Patent No. 9,796,833. 24 Oct. 2017.