

COST Action – FP1405 ActInPak – STSM report

Rheological characterisation of NFC/xylan-based hydrogels crosslinked by tannic acid and examination of aerogel structures with scanning electron microscopy

Mines ParisTech CEMEF – **Prof. Tatiana Budtova** (host)

University of Helsinki, Dept. of Food and Environmental Science – **Jose Martin Ramos Diaz** (visitor)

1. Introduction

This study was conducted in collaboration of two research institutions: Mines ParisTech CEMEF (Sophia Antipolis, France) and University of Helsinki (UH, Helsinki, Finland). Research groups in both institutions focus on the study of cellulose- and hemicellulose-based hydrogels and aerogels, which could have potential as novel biodegradable packaging material. Birch xylan is a plant cell wall polysaccharide (hemicellulose) that can be obtained from side-streams of the pulp paper industry; an strategic industrial sector in Finland. The potential of birch xylan as component in hydrogels and aerogels was studied in combination with nanofibrillated cellulose (NFC) and a food-grade cross-linking agent, tannic acid (TA, plant derived polyphenol). The utilization of effective though toxic cross-linking agents (e.g. ammonium zirconium carbonate) has greatly limited the development of food contact material. Preliminary experiments at UH showed that TA could be a promising cross-linking agent upon the formation of nanofibrillated cellulose (NFC)/xylan-based hydrogels and aerogels.

The aim of this study was to understand the interactions in the ternary system, NFC+xylan+TA, by conducting viscosity and rheological measurements. Additionally, the structure of aerogels was thoroughly examined with Scanning Electron microscopy (SEM).

2. Material and Methods

2.1. Materials

NFC (hydrogel, 2.65% aqueous dispersion) was purchased from UPM (Biofibrils AH80, Finland), and birch xylan (powder extract) was obtained from pressurized hot water extraction (PHME) of birch saw dust as described by Kilpeläinen et al. (2014). The TA (powder extract) used in this study was purchased from Sigma-Aldrich.

The aerogels were prepared by dissolving xylan in water under magnetic stirring at 80 °C. NFC and TA were added, and the suspensions were homogenized using an Ultra-Turrax (Ika-Werke, Staufen, Germany) at 1100 rpm for 15 min. The hydrogels obtained were cast into cubic Petri dishes (Sterilin Ltd., Newport, UK) and frozen for 24 h at -80 °C. The samples were then freeze-dried for 48 h.

2.2. Methods

Capillary viscometry

The viscosity of xylan solution, and xylan-TA suspension was measured through an Ubbelohde capillary viscometer (capillary type I, Lauda, Lauda-Koenigshofen, Germany) connected to an automatic pump (Lauda iVisc, Lauda, Germany). Pre-set temperatures (20, 35 and 50 °C) were controlled within ± 0.2 °C using a thermobath (Lauda E100, Lauda, Germany) with circulating water. The experimental error associated to the determination of relative viscosity was less than 2%.

Viscoelasticity of hydrogels

The storage and loss modulus of NFC-based hydrogels containing TA were analysed by a Bohlin GeminiTM 150 rheometer (Malvern Instruments, UK) equipped with a Peltier temperature control system maintained at 20 °C. A parallel plate fixture was used with a gap of 1 mm. A frequency test was conducted as a function of frequency on each sample from 0.1 to 10 Hz (descending frequency). All the tests were conducted within the linear viscoelastic region (moduli independent from the applied strain). Measurements were done in triplicate. Curves (corresponding to Eq. 1, 2) were fit to the experimental values of G' and G'' in order to examine structural properties numerically.

$$G' = G'_0 \times f^{n'} \quad (1)$$

$$G'' = G''_0 \times f^{n''} \quad (2)$$

Where G'_0 and G''_0 represent the deformation of G' and G'' ; the gap between G'_0 and G''_0 relate to the strength of intermolecular interactions of gel networks. Besides, n' and n'' indicate the rate of change of G'_0 and G''_0 with increasing frequency, f . According to Campo and Tovar et al. (2008), a stable gel should exhibit the same proportional change in G' and G'' with frequency over a wide range, thereby showing almost identical n' and n'' .

SEM

Experiments were performed using a scanning electron microscope PHILIPS JEOL 35CF (Hefts, UK) at the acceleration voltage of 10–15 kV. Prior to observation, thin layers of platinum (Metalizer MEB Quorum, Q150T ES, Quantum Tech Ltd., UK) were deposited by sputtering onto the surfaces of the aerogel samples (cross-section area).

3. Results

Viscosity measurement - xylan

The relative viscosity $[\eta_{relative} = \frac{t_{xylan\ solution}}{t_{distilled\ water}}]$ of xylan solutions increased in proportion to the concentration of xylan (**Figure 1, left**). However, only minor differences were observed at temperatures ranging from 20 to 50 °C. As expected, the intrinsic viscosity $[\frac{\eta_{relative}-1}{Xylan_{cc}}; Xylan_{cc} \rightarrow 0]$ of xylan solutions between 20 and 50 °C showed also negligible variation, from 0.0932 to 0.0954 (**Figure 1, right**). The inverse of the intrinsic viscosity at 20, 35 and 50 °C was 10.4, 10.7 and 10.5,

respectively. This means that, theoretically, xylan concentration should be at/above 11% to form a gel, regardless of the temperature.

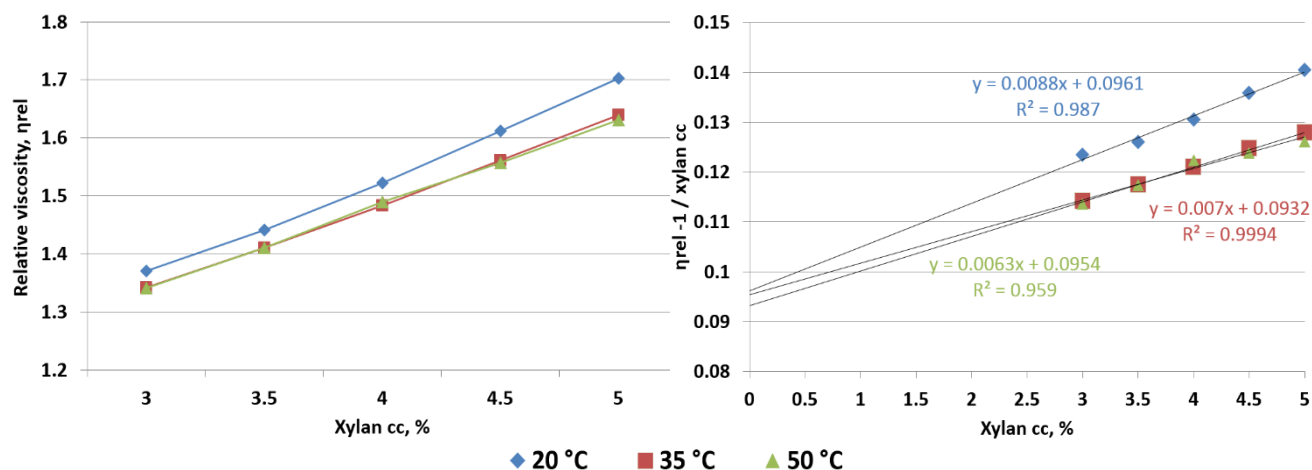


Figure 1. Relative viscosity (left) of aqueous solutions of xylan as a function of various xylan concentrations at 20, 35 and 50 °C. The intrinsic viscosity (left) was determined by extrapolating linear trendlines to zero xylan concentration, $\text{Xylan}_{cc} \rightarrow 0$. The corresponding equations are shown above.

Viscosity measurement – xylan and TA

The combination of xylan and TA (each one miscible in water) led to the formation of a suspension and, at larger concentrations, the formation of precipitate. The latter could be a major challenge upon the use of capillary viscometer. Therefore, preliminary tests were conducted to set a threshold of precipitation (**Table 1, Figure 2**). It was observed (visually) that the concentration of tannic acid could vary from 500 to 3000 ppm and that of xylan up to 1% in order to prevent the formation of heavy precipitate. This information was used to set working parameters.

Table 1. Testing of precipitate formation resulting from the combination of xylan and tannic acid.

Tannic acid, ppm	0.5% xylan	1% xylan
500	No precipitate	No precipitate
1000	No precipitate	No precipitate
1500	No precipitate	No precipitate
2000	No precipitate	No precipitate
3000	No precipitate	No precipitate
4000	No precipitate	Precipitate



Figure 2. Suspension (left) and precipitate (right) containing xylan and tannic acid.

The relative viscosity of various tannic acid concentrations (1000-3000 ppm) in 0.5% xylan (**Figure 3, left**) showed minor variation around 1 ($t_{TA+xylan} \cong t_{xylan}$). Similarly, the relative viscosity of various xylan concentrations in 3000 ppm TA showed also negligible variation around 1. Given the resulting data, calculations of intrinsic viscosity were considered irrelevant.

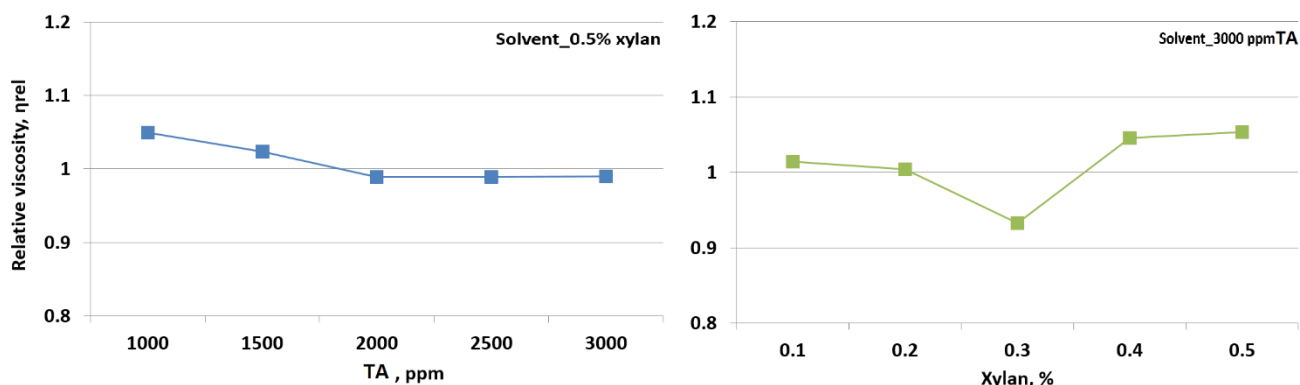


Figure 3. Relative viscosity of tannic acid-0.5% xylan (left) and xylan-3000 ppm tannic acid (TA, right) solutions as a function of various TA and xylan concentrations, respectively. $T=20\text{ }^{\circ}\text{C}$.

Viscoelastic properties of hydrogels – NFC and TA

The incorporation of higher contents of TA into NFC-based hydrogels seemed to increase G'_0 , G''_0 and the gap between G'_0 and G''_0 , up to a level comparable to that of the reference sample (without TA) (**Figure 4, left**). Additionally, the gap between n' and n'' reduced as the content of TA increased (**Figure 4, right**). This could be interpreted as greater structural stability given the ideal: $|n'-n''| = 0$.

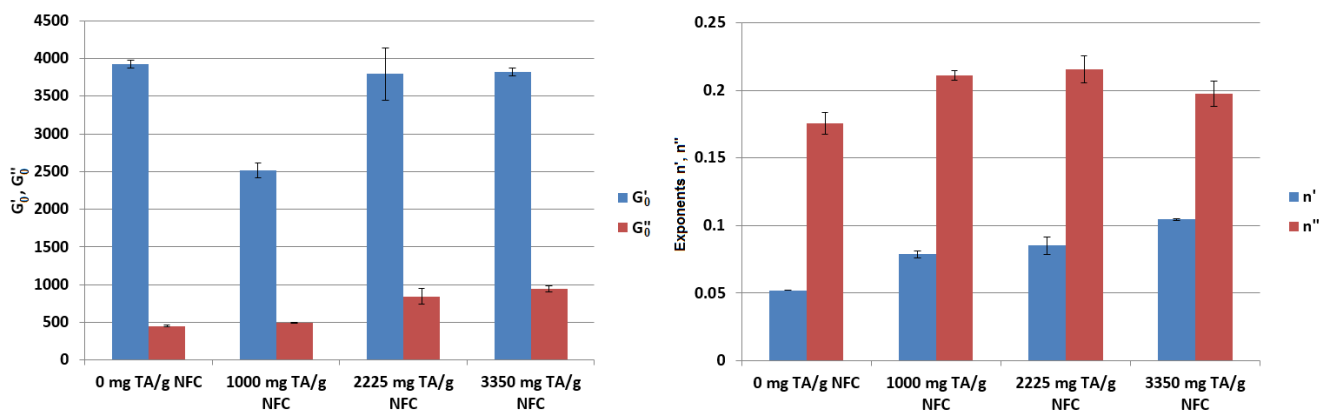


Figure 4. Fitted values of G'_0/G''_0 and n'/n'' for NFC-based hydrogels containing various contents of tannic acid.

Viscoelastic properties of hydrogels – Xylan

As indicated in the section 'viscosity measurement – xylan', xylan should (in theory) be able to form a gel-like system if its concentration in water is equal or above 11%. In this particular test, the higher concentration of xylan (2.5 – 15%) showed no proportionality with increasing G' and gap between G' and G'' (**Figure 5, left**). Besides, large standard deviations indicate a great deal of randomness in the data (at least, under the conditions tested). Regarding n' and n'' as indicators of the structural stability,

samples with higher concentration of xylan showed similar values of n' and n'' and lower standard deviation compared to that of distilled water and 2.5% xylan.

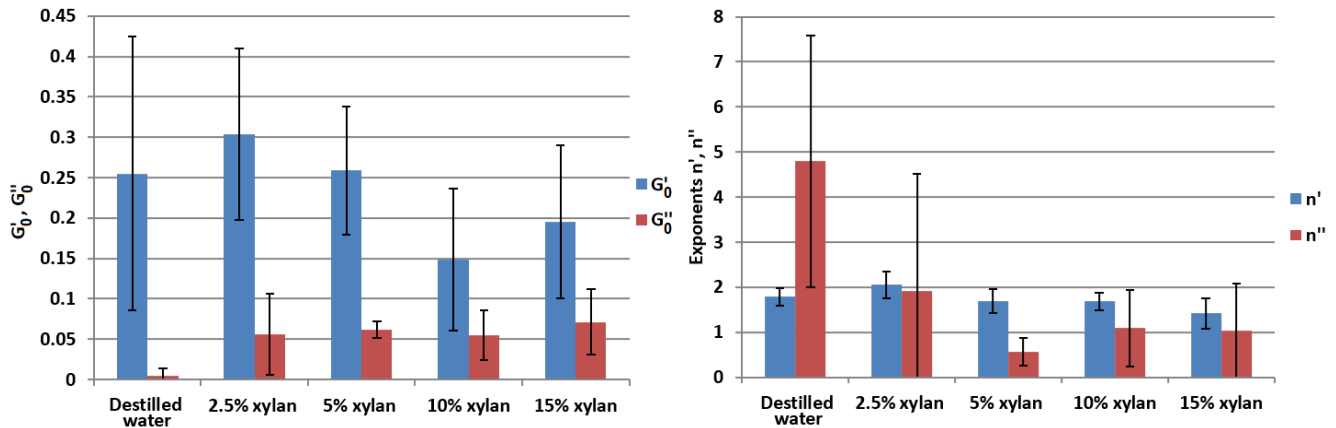


Figure 5. Fitted values of G'_0/G''_0 and n'/n'' for aqueous solutions containing various concentrations of birch xylan.

Electron microscopy

Differences were observed between aerogels containing 1.5% total polysaccharide (40% xylan, 60% NFC) with and without TA. In general, aerogels with TA exhibited smoother surfaces, more homogeneous porous structures and thinner walls than those without TA; similar features were observed under optical microscopy at University of Helsinki. At greater magnification (2000x and 10000x), xylan and NFC were distinguishable components in aerogels without TA (**Figure 6, A and C**) while, in aerogels with TA, it was difficult to discriminate between xylan and NFC (**Figure 6, B and D**).

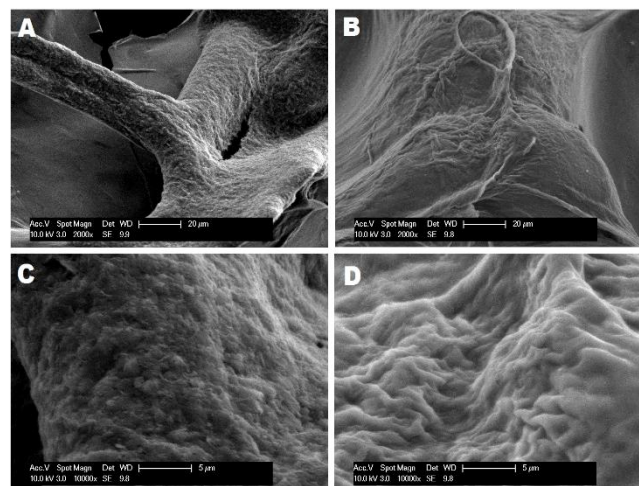


Figure 6. Scanning electron micrograph of xylan-NFC-based aerogel samples. A and C, 1.5% Total polysaccharide (40% xylan, 60% NFC) without TA. B and D, 1.5 % Total polysaccharide (40% xylan, 60% NFC) with TA.

Aerogels containing 3.5% total polysaccharide (70% xylan, 30% NFC) were observed to have similar structures regardless of the presence of TA. At greater magnification, xylan and NFC were easily discriminated in aerogels without TA (**Figure 7, A and C**) while the opposite happened in the presence of TA (**Figure 7, B and D**). The beads found in aerogels with TA most probably respond to

the complexation of xylan and TA. Similar beads were not observed in aerogels containing 1.5% total polysaccharides with TA.

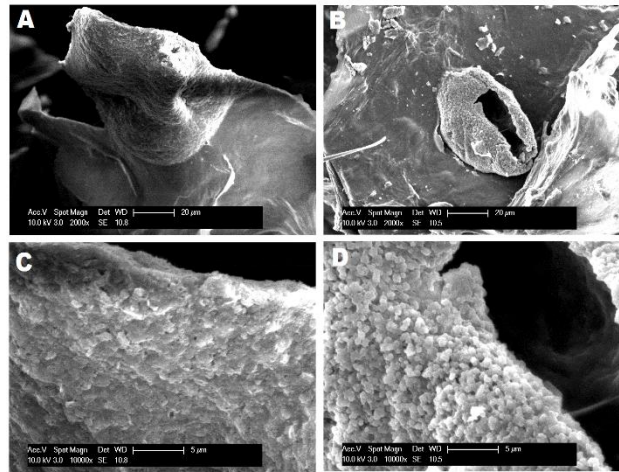


Figure 7. Scanning electron micrograph of xylan-NFC-based aerogel samples. A and C, 3.5% Total polysaccharide (70% xylan, 30% NFC) without TA. B and D, 3.5 % Total polysaccharide (70% xylan, 30% NFC) with TA.

4. Publication plan

The information presented in this report will be included in a manuscript for publication in a peer-reviewed journal. Besides, the results will be presented during the next ActinPak meeting in 2017.

5. Conclusions

There is considerable evidence suggesting that xylan and TA form complexes thereby altering the type of interaction with NFC. The formation of suspensions and inability to increase viscosity in aqueous medium indicate that xylan and TA, rather than being dispersed, form strong bonds at immediate contact. The sharp differences in the morphology of aerogels with or without TA gave hints on the degree of interaction between xylan-TA complexes and NFC. Besides, TA alone might have an effect on hydrogel stability, but this requires further testing. Regarding xylan alone, its ability to form hydrogels could not be proven in the present study, yet this information should also be reconfirmed with instruments of greater sensitivity (e.g. interfacial shear rheometer).

This is an important step in the development of aerogels as food contact material as xylan-TA complexes seem to form stronger bonds with NFC thereby increasing the structural strength of hydrogels and, particularly, aerogels. This clearly supports the physical measurements conducted at UH during 2016.

6. Reference

- Kilpelainen, P., Leppänen, K., Spetz, P., Kitunen, V., Ilvesniemi, H., Pranovich, A., Willför, S. 2012. Pressurised hot water extraction of acetylated xylan from birch sawdust. *Nordic Pulp and Paper Research Journal*, 27, 1-8.
- Campo, L., Tovar, C. 2008. Influence of the starch content in the viscoelastic properties of surimi gels. *Journal of food engineering*, 84, 140-147.