DNA/Cellulose hybrid nanomaterials

A. Mangalam, J. Simonsen and A.S. Benight

Oregon State University
120 Richardson Hall, Corvallis, OR, 97331, USA
john.simonsen@oregonstate.edu

ABSTRACT

One potential advantage of nanotechnology is the ability to engineer bottom-up hierarchical assembly of nanoparticles into functional macroscale materials and devices. The most promising mechanism for accomplishing this is self-assembly. In this project, an advanced strategy was developed to design and manufacture novel hierarchical nanomaterials from cellulose nanocrystals, a molecular coupler and DNA. Recently, cellulose nanocrystals (CNXLs) have attracted much attention from researchers for their remarkable reinforcing abilities. Some advantageous properties of CNXLs are their high aspect ratio of around 20-50 (length/width), low density of around 1.56 g/cc, high stiffness and strength reported to be 145 GPa and 7500 MPa respectively. DNA and coupler were successfully grafted to cellulose nanocrystals. The size, shape and material properties of the resulting hybrid nanomaterial were investigated via UV spectroscopy, NMR, dynamic laser light scattering, thermal analysis and mechanical testing.

Keywords: DNA, cellulose, nanoparticle, nanocrystal, composite

1 INTRODUCTION

Cellulose nanocrystal (CNXL)/polymer composites have attracted the attention of several researchers in the past few years. These nanocomposites have drawn special attention from researchers because of their remarkable reinforcing capability, excellent mechanical properties, low density and ability to form composites with thermoplastic matrices. However, the limitations of reinforcement theory apply to nanoparticles as well as conventional filler particles, thus understanding and controlling filler-matrix interactions are necessary in order to fully exploit these materials. In this project we take a novel approach and construct fully cross-linked system composed of CNXLs and DNA oligomers. This material should be self-assembling via duplex formation of the DNA oligomers. Since all the components are either covalently bonded or bonded through duplex formation, there is no interface, or interphase, between the filler and matrix. We herein report on the initial work of grafting the DNA oligomers to the CNXL surface as the first phase in this larger process.

A short single stranded oligonucleotide with 78 DNA bases with a dodecylamine modifier grafted to the 5' end were grafted on the modified CNXL surface using the carbodiimide coupling reaction. A second set of complementary oligonucleotide was grafted on CNXL. The DNA-grafted CNXLs formed stable suspensions in water. When mixed under suitable conditions, duplex formation was evident and followed by means of dynamic laser light scattering.

2 EXPERIMENTAL

2.1 CNXL preparation and TEMPO mediated oxidation

Cellulose nanocrystals (CNXL) were prepared by sulfuric acid hydrolysis of cotton cellulose using well documented methods[1]. The resulting CNXL dispersion (in water) was then subjected to oxidation by converting primary hydroxyls to carboxylic acids by means of 2,2,6,6-tetramethylpiperidine-1-oxyl radical (TEMPO) mediated oxidation [2-4]. The resulting carboxylated CNXLs were then characterized by FTIR (Thermo Nicolet-Nexus 470) to confirm the presence of COOH group on its surface. The carboxyl content on the CNXL surface was also quantified by potentiometric acid-base titration.

2.2 Oligonucleotide grafting on CNXL

A 78-mer DNA oligomer with a dodecyl amine group grafted to the 5' end was purchased from IDT Corp, Coralville, Iowa. The amine group was grafted to the carboxylic group on the modified CNXLs using 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (EDC) coupling [5-8]. The reaction mixture was cleaned using ultrafiltration in order to remove unreacted EDC and oligomer from the reaction mixture. The sample was then subjected to UV-Vis analysis. Absorbance of DNA at 258 nm was used to determine the presence and extent of grafting.

3 RESULTS

3.1 Carboxylation of CNXL by TEMPO mediated oxidation.

The presence of carboxylic acid groups on the CNXL surface was confirmed by FTIR. A small peak at 1739 cm-1 and 1635 cm-1 indicated the presence of free carboxylic acid and aldehyde groups respectively on the pure CNXL surface (Fig. 1). This may be due to the oxidation of hydroxyl groups by sulfuric acid during the initial hydrolysis of the cotton. The presence of the carboxylic acid group was confirmed by the strong signal at 1733.5 cm-1 (Fig. 2). A potentiometric titration performed to quantify the carboxylate content gave values typically between 0.95 to 1.3 mmol CO2H/g CNXL depending on the reaction conditions and oxidation time.

3.2 Oligonucleotide grafting to CNXL using EDC

The grafting of the amino group on the 5' end of C12 chain of the oligonucleotide on the carboxylic group on CNXL was determined using UV-Vis analysis. The presence of unreacted oligomer was followed by measuring the UV absorbance of the reaction mixtue after successive rinsing in a 50 nm pore size ultrafilter.

UV absorbance at 258 nm of the oligonucleotide grafted CNXL and control were plotted against the number of cleaning cycles performed (Figure 3).

The absorbance showed a gradual drop from 3.9 to 2 during successive cleanings (1st to 6th) and then remained constant. The control showed a sharp decrease in the absorbance from 3.9 to 0.5 after the first cleaning and then remained almost stable. This clearly implied the absence of linkage between cellulose and oligonucleotide in the control and the successful grafting of the DNA oligomer to the CNXL surface.

An FTIR analysis of carboxylated and oligonucleotides-grafted CNXL confirmed the successful graft (Figure 4). The clear shift in the carbonyl peak from 1730 cm-1 to 1640 cm-1 suggests that free carboxylic acid groups on the surface were converted to amide groups due to the EDC coupling reaction.

REFERENCES

[1] Orts, W J.; Imam, S H.; Guttman, M. E.; Nguyen, A. Annual Technical Conference - Society of Plastics Engineers 2004, 62, 2427.

[2] Bragd, P., L.; Besemer, A. C.; van Bekkum, H. Journal of Molecular Catalysis A: Chemical 2001, 170, 35.

[3] de Nooy, A. E.; Besemer, A. C.; van Bekkum, H. Carbohydrate research 1995, 269, 89.

[4] Besemer, A. C.; de Nooy, A. E. J.; van Bekkum, H. 1996.

[5] Ficht, S.; Roglin, L.; Ziehe, M.; Breyer, D.; Seiz, O. Synlett 2004, 14, 2525.

[6] Lloyd, R. D.; Burns, M. C. Journal of polymer science 1979, 17, 3459.

[7] Araki, J.; Wada, M.; Kuga, S. Langmuir 2000, 17, 21.

[8] Hermanson, T. G. Zero length crosslinkers. In Bioconjugate techniques.; Academic press: Rockford, Illinois., 1996; pp 170.

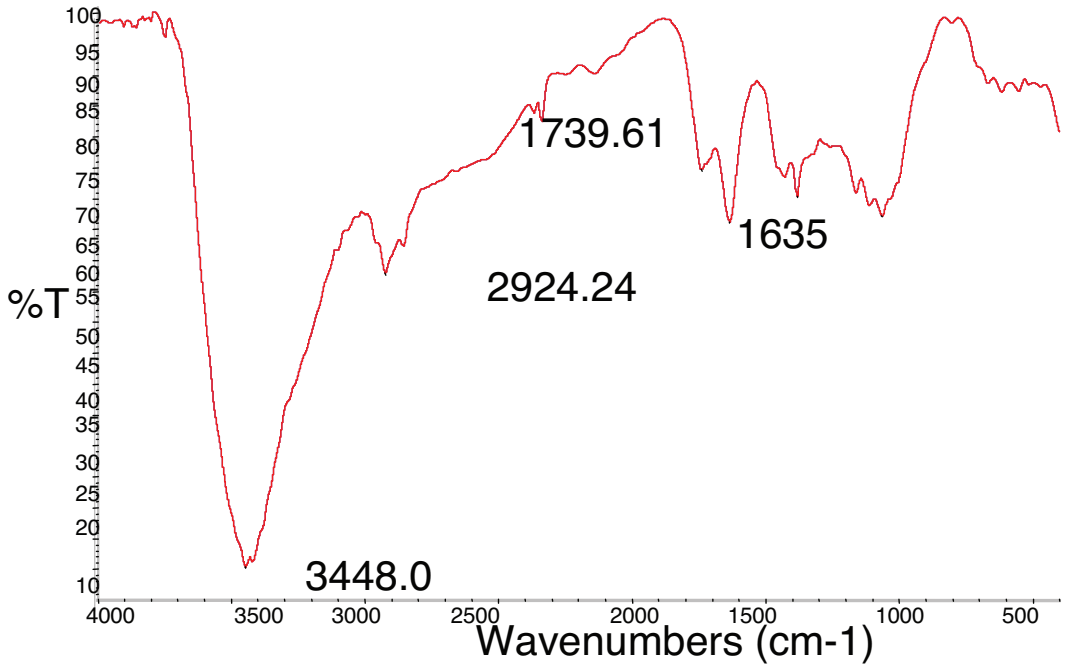

Figure 1: FTIR of CNXL.

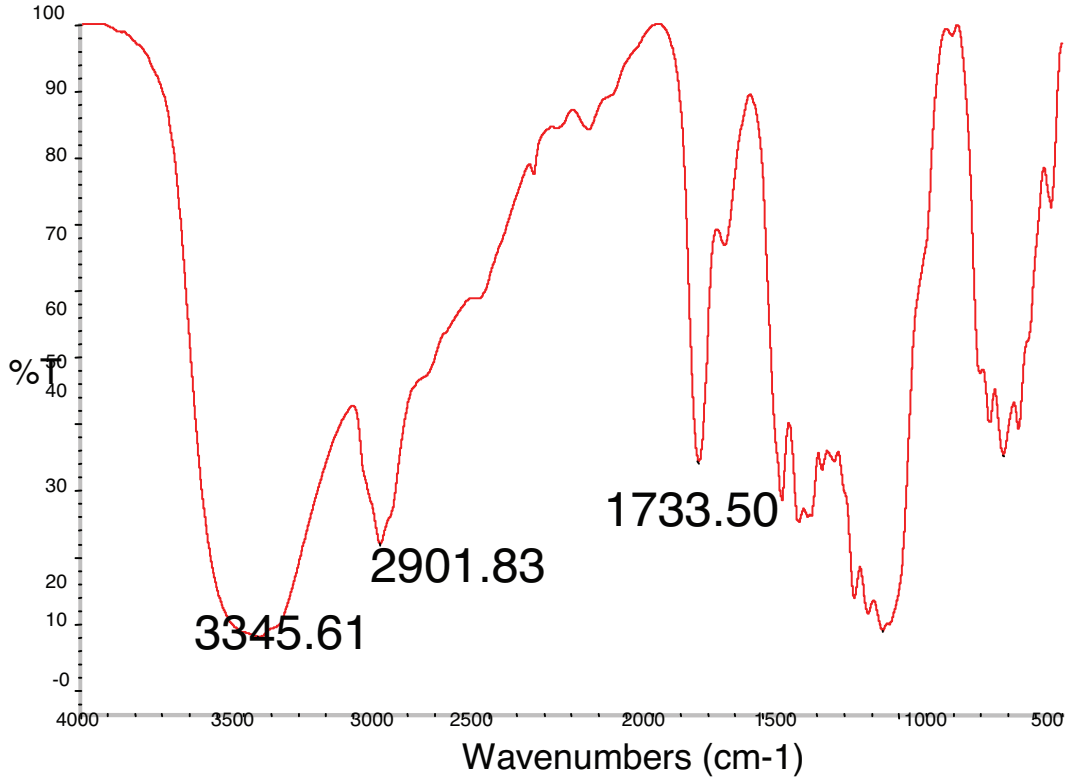

Figure 2: FTIR of carboxylated CNXL

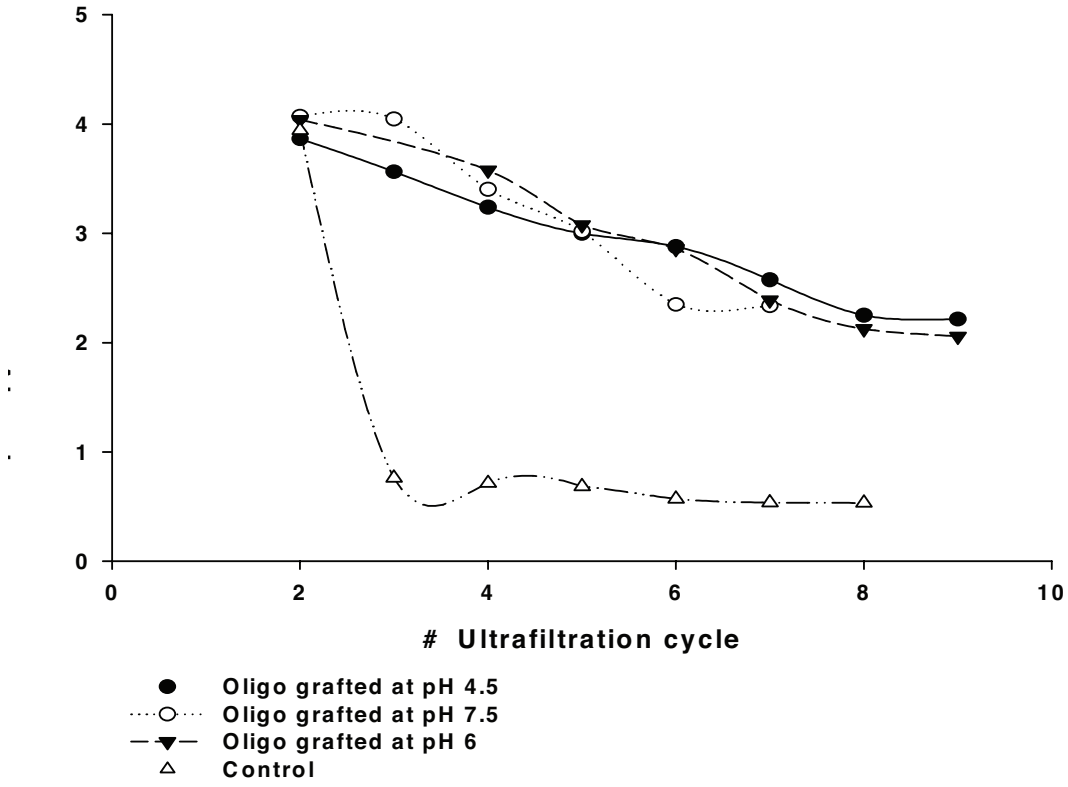

Figure 3. Plot showing UV absorbance of DNA oligomer grafted on CNXL after each cleaning cycle.

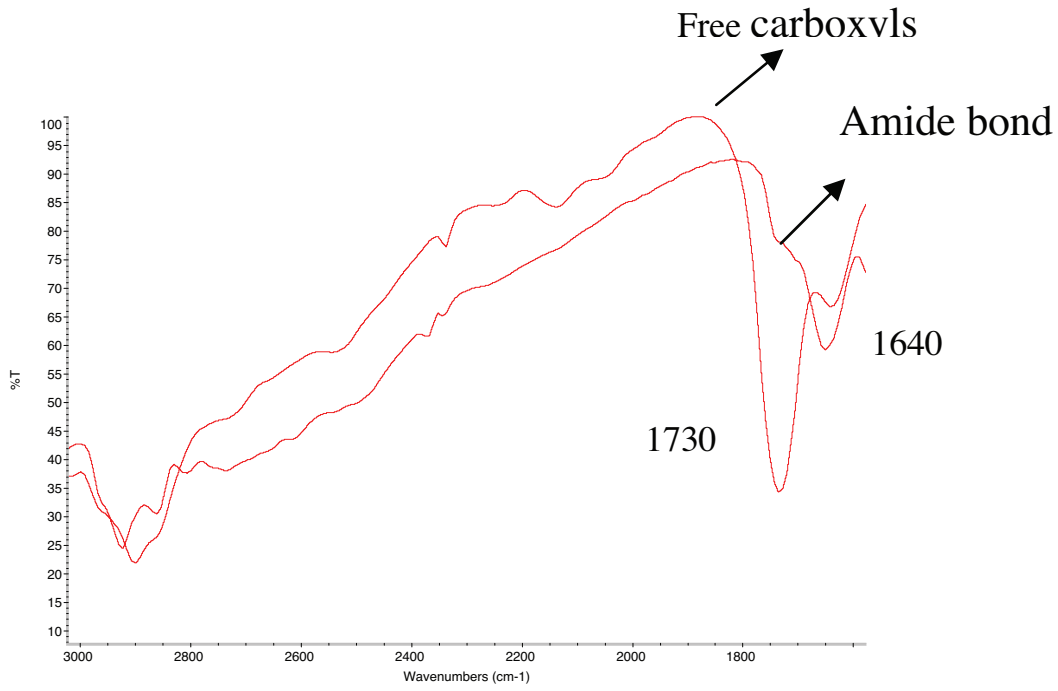

Figure 4: FTIR spectra of carboxylated and oligomer grafted CNXLs.