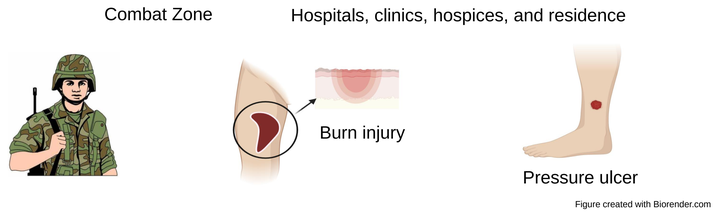
APPLICATION OF HYDROGEL CELLULOSE NANOFIBER-BASED COMBINATION SCAFFOLDS IN CUTANEOUS WOUND HEALING

J. Vincent Edwards[1], Nicolette Prevost[1], Michael Easson[1], Jacobs Jordan[1], Katie Hamel[2], Haley Lassiter[2], Emma Rogers[2], Jordan Robinson[2], Trivia Frazier[2], Cecilia Sanchez[2]
[1]U.S. Department of Agriculture, ARS, Southern Regional Research Center, New Orleans, LA, [2] Obatala Sciences Inc., New Orleans, LA

Introduction

Cellulose nanofibers (CNF) offer promising potential for wound healing and tissue engineering due to their non-immunogenic, low-protein absorbing, biocompatible nature and high specific surface area. Our study aimed to explore the feasibility of using CNF/human-derived hydrogel combinations (NHC) as scaffolds for wound healing, particularly in burn injuries and chronic wounds. We hypothesized that the combination gels could maintain structural, healing, and antibacterial properties to promote wound healing.



Methods and Concept

The study involved the biophysical characterization and assessment of the biocompatibility and wound healing capacity of two of two forms of CNF (CNF and TEMPO) in combination with human-derived hydrogels. For characterization, rheology studies, evaluation of gelation time, protein release studies, proteomics, and microstructure analysis via SEM were performed. The biocompatibility of the nanocellulose/hydrogel combinations was assessed via proliferative and adipogenic differentiation capacity of human adipose-derived stromal/stem cells (ASCs) and human dermal fibroblasts (DFCs). Finally, wound healing studies were performed with ASCs.

Conclusions and Future Directions

Cotton-derived CNF and TEMPO **a)** provide structural support and integrity to human-derived hydrogels; **b)** increase viscosity and hydrogel stiffness in combination with human-derived hydrogels; **c)** are biocompatible by supporting DFC proliferation and ASC adipogenic differentiation.

Further studies are needed to identify wound healing properties in wound healing animal models as well as to identify antibacterial activities.

Acknowledgments
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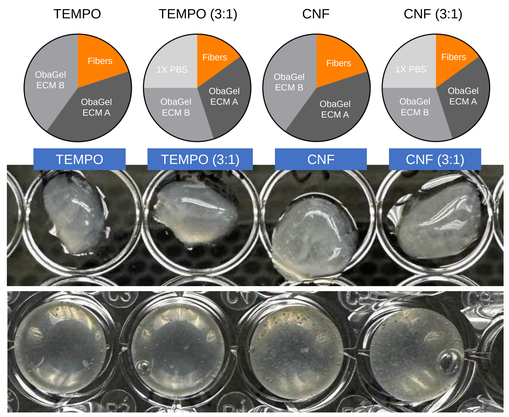
- Dr. Zhang of the Ohio State University CCIC for proteomics
- Dr. Jibao of Tulane University for SEM imaging

References
- Belgodere, JA et al. (2023); Bender et al. (2020).

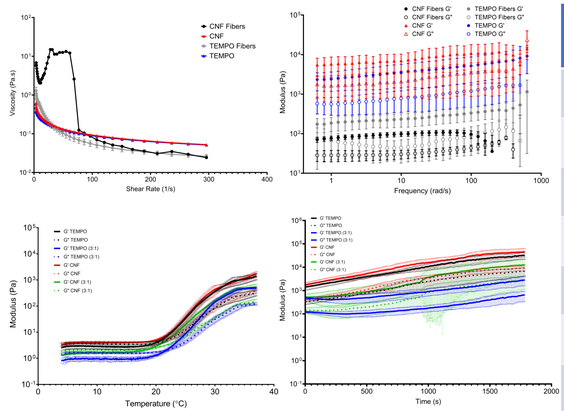
Contacts
- Vince Edwards: vince.edwards@usda.gov
- Cecilia Sanchez: Cecilia.sanchez@obatalasciences.com

Results



Mixture	% W/V Nanofibers
TEMPO Fibers	10%
TEMPO	2%
TEMPO (3:1)	1.5%
CNF Fibers	10%
CNF	2%
CNF (3:1)	1.5%

Figure 1. Hydrogel Composition and Proteomics. Hydrogel/cotton-based scaffolds were prepared using a combination of TEMPO and CNF cotton-derived nanofibers with Obatala Sciences' human-derived ObaGel®-ECM hydrogel. Gelation of the combinations was visually assessed after 30 min incubation at 37°C.



Mixture	Storage Modulus (G', Pa)	Loss Modulus (G", Pa)	Viscosity (Pa*s)
TEMPO Fibers	220.60	43.17	0.0562
TEMPO	3309.77	726.36	0.0899
TEMPO (3:1)	3212.47	602.73	n/a
CNF Fibers	94.23	29.24	0.0867
CNF	6806.11	1976.14	0.0945
CNF (3:1)	1337.29	284.00	n/a
ObaGel ECM	64193.1	8131.07	0.0669

Figure 2. Rheological Characterization. CNF and TEMPO formulations exhibited similar viscosity and stable, well cross-linked profiles with frequency interrogation. All solutions crosslinked at 37°C. The diluted CNF and TEMPO mixtures demonstrated less stiffness compared to the non-diluted mixtures. The TEMPO solutions displayed a greater stiffness compared to the CNF solutions.

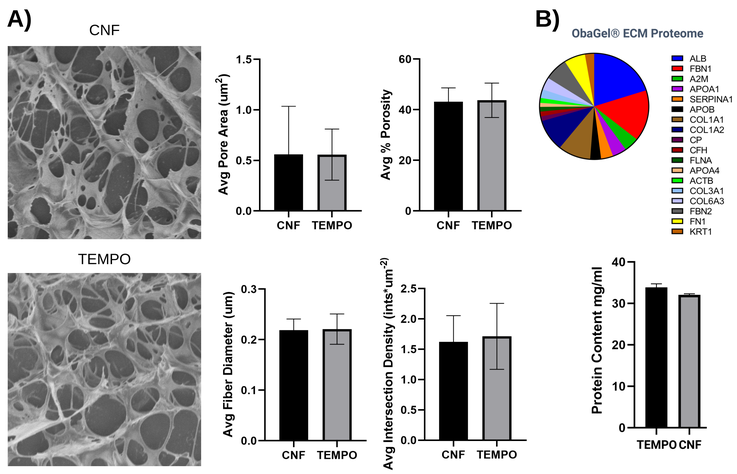


Figure 3. SEM Analysis. A) SEM imaging and analysis revealed that both the CNF and TEMPO hydrogel mixtures exhibit similar microstructures. **B)** The ObaGel®-ECM proteome profile demonstrated an extracellular matrix protein dominant composition. BCA assay results revealed similar protein contents for both the TEMPO and CNF hydrogel mixtures.

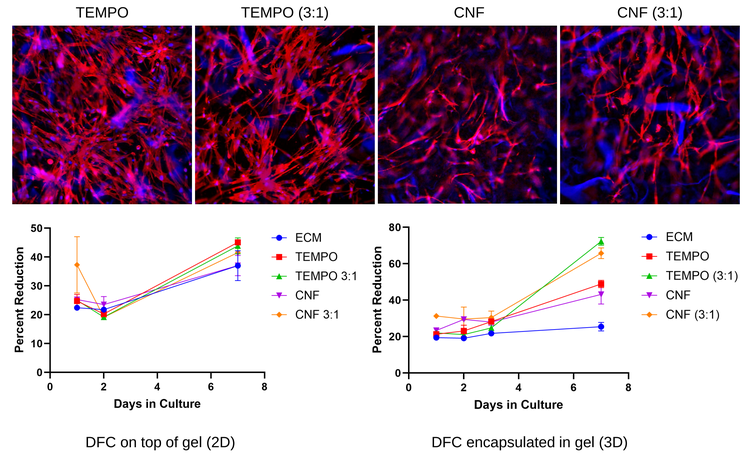


Figure 4. Biocompatibility. DFCs cultured on the surface or encapsulated in the CNF and TEMPO hydrogels demonstrated proliferative capacity and increased metabolic activity as represented by percent reduction of alamarBlue. (red=cytoskeleton, blue=nuclei)

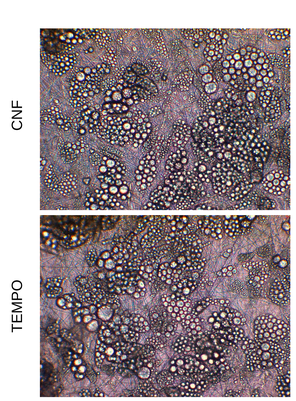


Figure 5. Hydrogels Support Adipogenesis. ASCs exhibited similar lipid deposition on TEMPO and CNF hydrogels.

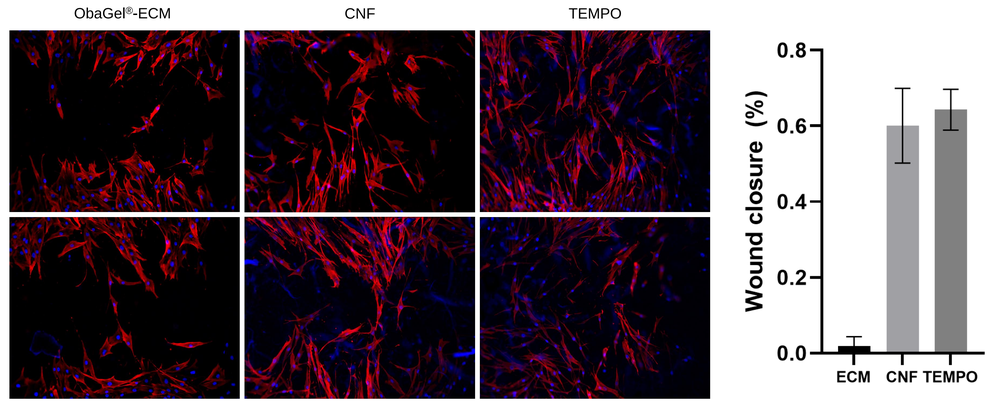


Figure 6. Wound Healing Assay. ASC migratory behavior is more profound on the TEMPO hydrogel compared to CNF and ObaGel®-ECM B. (red=cytoskeleton, blue=nuclei)