Automated Nucleic Acid Isolation from Nanocellulose-Based 3D Cell Cultures

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Aim of the study

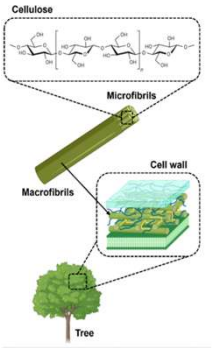
Nanofibrillated cellulose (NFC) is a bio-based nanomaterial capable of forming hydrogels [1]. Due to the porous and soft structure, hydrogels are well suitable for 3D cell culture applications, mimicing in vivo conditions more accurately than conventional 2D cell culture on flat surfaces [2].

In this study, the BeadHunter™ automated platform for nucleic acid isolation was evaluated, with particular focus on its performance in NFC-based 3D cell culture models.

BeadHunter™ Automated Platform

Nanofibrillated Cellulose (NFC)

Nanofibrillated Cellulose



Results

Comparison of Isolation Methods

The performance of BeadHunter was compared to manual isolation and the KingFisher platform in 2D cell cultures. The manual method provided the highest yield, but the sample variability was high.

The automated methods demonstrated higher reproducibility, and the BeadHunter platform showed competitive performance to manual and KingFisher platform methods.

NFC Hydrogel Rheological Characteristics

Nanofibrillated cellulose forms a hydrogel, whose viscoelastic properties are characterized by the storage modulus (G', elasticity) and the loss modulus (G", viscosity). The results showed that G' > G", indicating that elastic behavior predominates.

The increase in both moduli as a function of frequency suggests the presence of a physically cross-linked hydrogel network structure.

Nucleic Acid Extraction from 3D Cultures

RNA yield and purity were compared between 2D and NFC-based 3D cell culture models. The RNA concentration in the 3D models was lower than that observed in the 2D samples.

Although the 260/280 ratios indicated high purity, the lower 260/230 ratios in the 3D samples suggest the presence of impurities. These results indicate that the hydrogel matrix reduces the efficiency of nucleic acid isolation.

Comparison of Isolation Methods

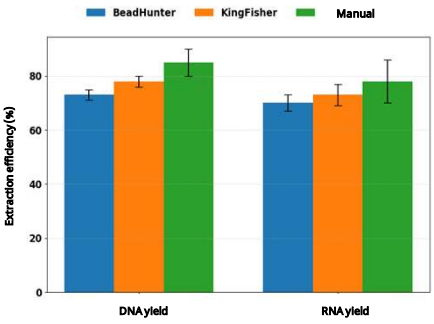


Figure 1. DNA- ja RNA-yields with different extraction methods using 2D cell model.

Rheological Analysis

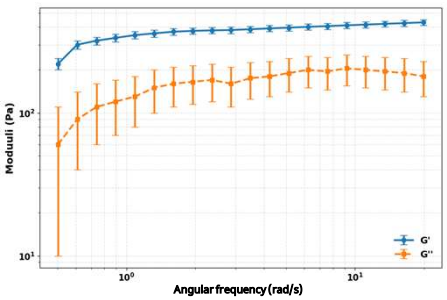


Figure 2. NFC Rheology: Storage modulus (G') and loss modulus (G'') as a function of angular frequency.

RNA-extraction from 2D/3D- Cell Models

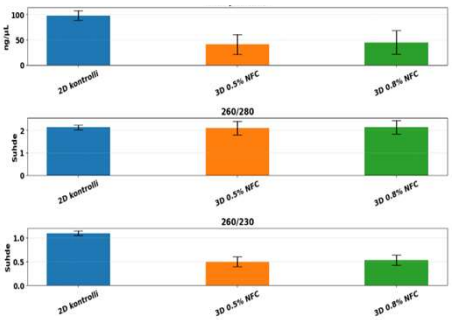


Figure 3. RNA concentrations and 260/280 and 260/230 ratios in 2D and 3D cell culture models.

Conclusion

Automated BeadHunter extraction method demonstrated competitive performance compared to other isolation methods. However, the RNA yield obtained with the NFC-based 3D models was lower and requires further optimization.

Reference

[1] Klemm D, Kramer F, Moritz S, Lindström T, Ankerfors M, Gray D, Dorris A. Nanocelluloses: a new family of nature-based materials. Angew Chem Int Ed Engl. 2011 Jun 6;50(24):5438-66.

[2] Lou YR, Leung AW. Next generation organoids for biomedical research and applications. Biotechnol Adv. 2018 Jan-Feb;36(1):132-149.

Research Collaborators

The nanofibrillated cellulose in the study was developed in an international collaboration between the Biopharmaceutics Research Group at University of Helsinki and Khalifa University.

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