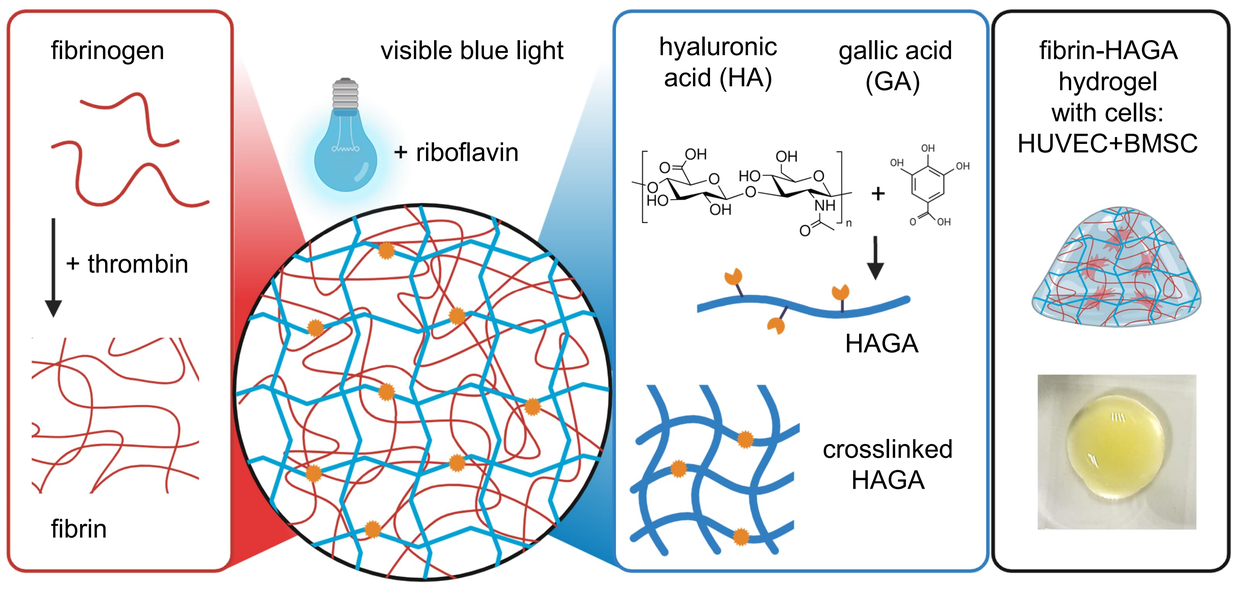
Tampere University

Centre of Excellence in **Body-on-Chip** Research

Adult Stem Cells

Novel Blue Light-Crosslinked Fibrin-Hyaluronan Composite Hydrogel for Vascularized Tissue Engineering

Arjen Gebraad[1,2,3], Anastasiia Mykuliak[1,2], Austin D. Evans[4], Antti Eskelinen[5,6], Kirsi Kuismanen[7], Oommen P. Oommen[4,8], Susanna Miettinen[1,2]



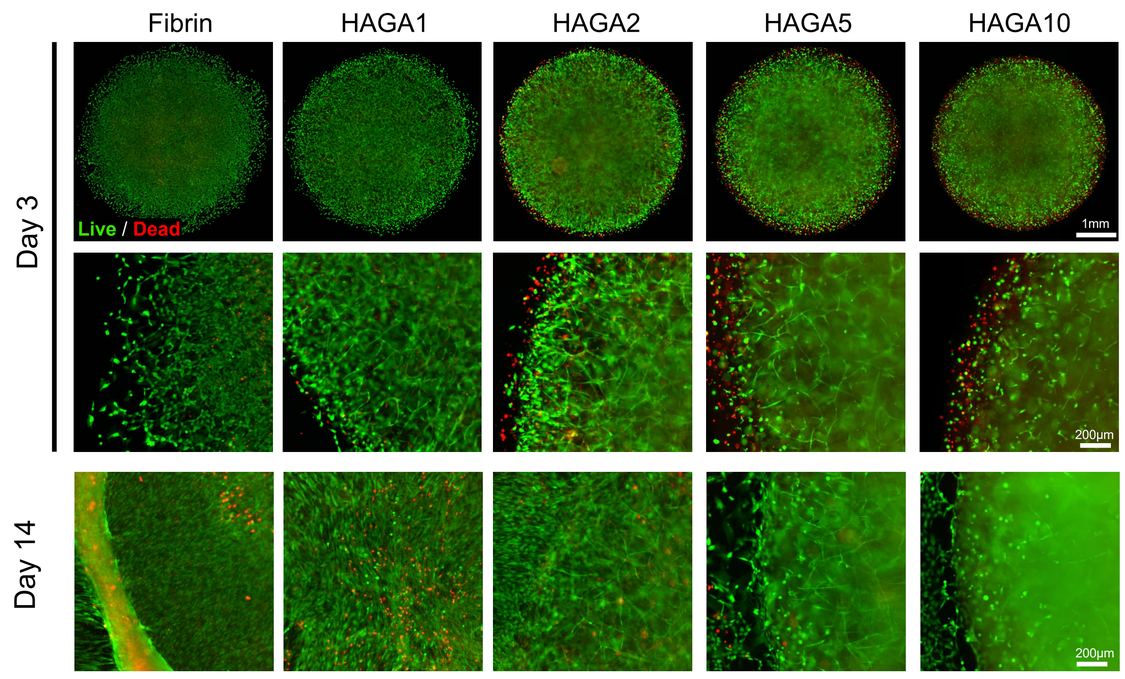
Background

Fibrin is widely used in tissue engineering and microphysiological systems because of its excellent biocompatibility and user-friendliness. Its intrinsic pro-angiogenic properties are critical for the development of vascularized tissue constructs, facilitating nutrient and oxygen delivery and enhancing physiological relevance. However, the limited mechanical strength of fibrin often results in rapid hydrogel contraction and degradation. In this study, we investigated a novel blue light-crosslinked fibrin-hyaluronan hydrogel as an effective strategy to enhance the long-term stability of fibrin hydrogel and evaluate its potential for vascular tissue engineering and vascularized organ-on-chip applications.

Materials and methods

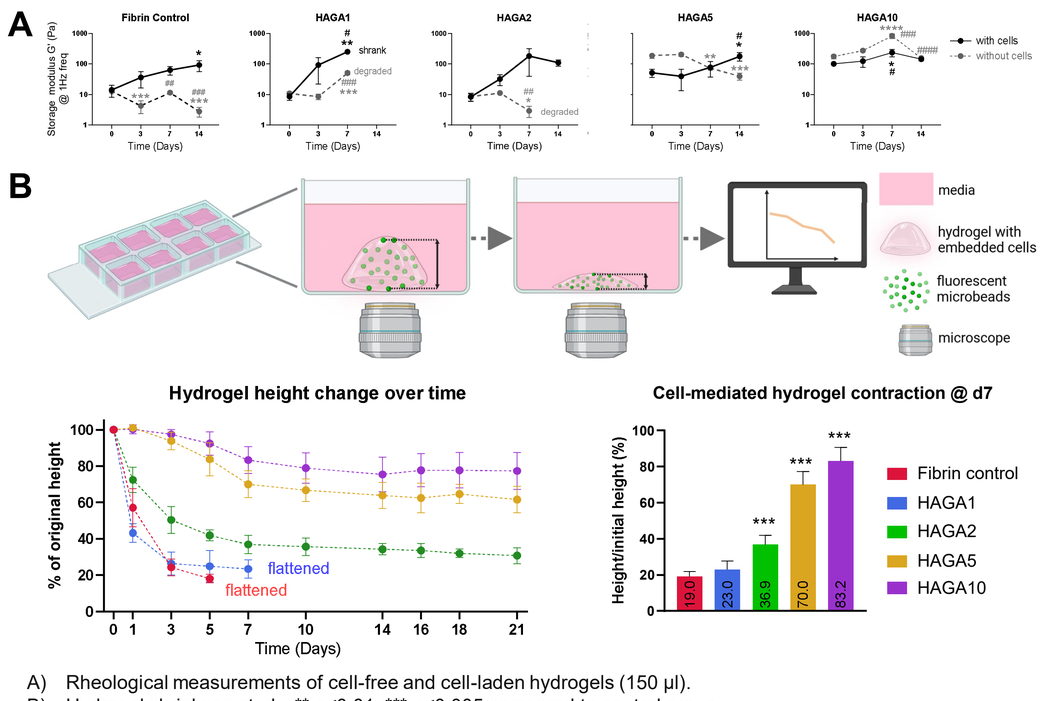
Microvascular networks were formed by human umbilical vein endothelial cells (HUVECs) and bone marrow-derived mesenchymal stem/stromal cells (BMSCs) within hydrogels composed of 2.5 mg/ml fibrin and blue light-crosslinked gallic acid-functionalized hyaluronic acid (HAGA; 1, 2, 5, and 10 mg/ml final concentration). Hydrogel compaction, encapsulated cell viability and proliferation were monitored for three weeks and compared to pure fibrin hydrogel. Microvascular network area coverage, total vasculature length, and pericyte alignment were assessed after one week of culture. Additionally, microvascular networks were established within AimBiotech IdenTx microfluidic chips.

1 Good cell viability and proliferation



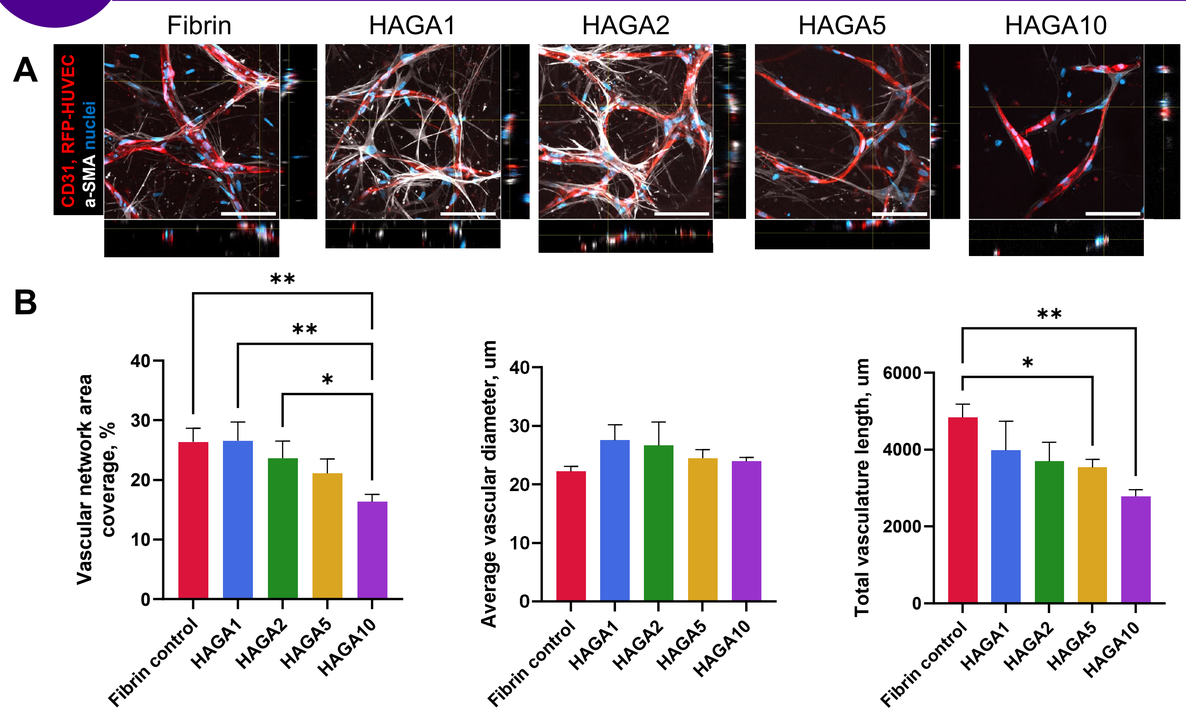
Live/dead staining to assess cell viability of HUVEC-BMSC co-cultures encapsulated in hydrogels (10 µl).

2 Improved mechanical properties, shrinkage resistence → extended culture time



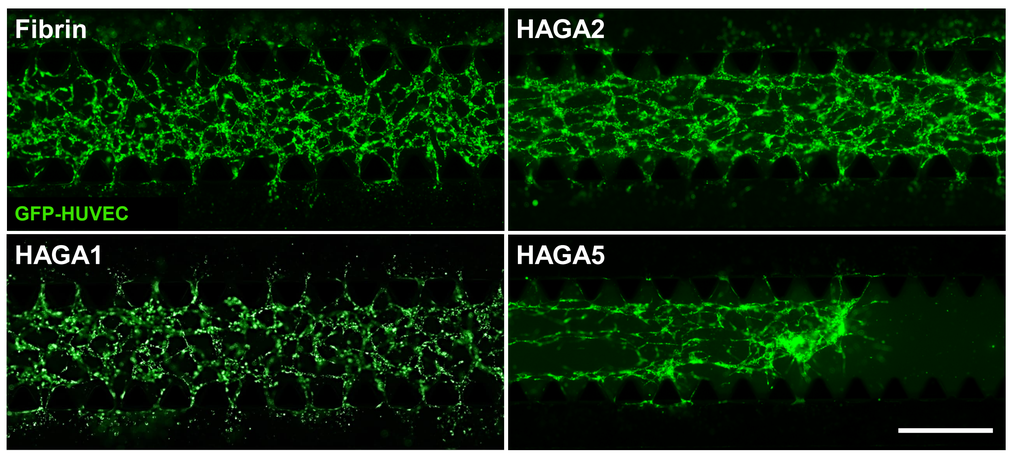
A) Rheological measurements of cell-free and cell-laden hydrogels (150 µl).
B) Hydrogel shrinkage study. ** p≤0.01, *** p≤0.005 compared to control group

3 Microvascular Network Formation



A) Confocal images showing α-SMA positive BMSCs in close proximity to RFP-HUVEC vessels after 7 days of culture. Scale bars: 100 µm.
B) Quantified morphological characteristics of the microvascular networks. *p<0.05, **p<0.01.

4 Vasculogenesis on-chip



Microvascular networks formed in IdenTx microfluidic chip by co-culture of GFP-HUVECs and BMSCs for 7 days
Total cell concentration: 12 million cells/mL. Scale bar: 1 mm.

Conclusions

A novel blue-light crosslinked fibrin-HAGA hydrogel is a promising candidate for vascularized tissue engineering and organ-on-a-chip applications. Hydrogel formulations of fibrin with 2 mg/ml and 5 mg/ml HAGA showed the best results in terms of shrinkage resistance, long-term culture, and vascular network formation.

Affiliations: [1] Adult Stem Cell Group, Faculty of Medicine and Health Technology, Tampere University, Tampere, Finland; [2] Tays Research Services, Wellbeing Services County of Pirkanmaa, Tampere University Hospital, Tampere, Finland; [3] Biomedical Spectroscopy Laboratory, Department of Technical Physics, University of Eastern Finland, Kuopio, Finland; [4] Bioengineering and Nanomedicine Group, Faculty of Medicine and Health Technology, Tampere University; [5] Faculty of Medicine and Health Technology, Tampere University; [6] Coxa Hospital for Joint Replacement, Tampere, Finland; [7] Department of Obstetrics and Gynecology, Tampere University Hospital; [8] School of Pharmacy and Pharmaceutical Sciences, Cardiff University, UK

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CENTRES OF EXCELLENCE IN RESEARCH

SUOMEN KULTTUURIRAHASTO

Finnish Cultural Foundation

Experimental work, analyses and graphics:
Anastasiia Mykuliak, MSc

Contact:
Arjen Gebraad, PhD
arjen.gebraad@tuni.fi