**Development of a 3Rs-Based Synovium-on-Chip Platform for Modelling Rheumatoid Arthritis and Predicting Treatment Response**

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**Background and Aims**

Rheumatoid arthritis (RA) is a chronic autoimmune disease primarily affecting the synovial joints, with a global prevalence of ~0.5–1%. Despite numerous therapeutic options, over 40% of patients experience long-term disability due to the absence of predictive tools for treatment response. To address this gap, we developed a **patient-specific synovium-on-chip (SoC)** platform using RA cells from minimally invasive biopsies. This 3Rs-compliant model enables personalized clinical trials-on-chip to predict drug efficacy.

**Methods**

A 3D microfluidic platform was designed to host autologous, leukocyte-infiltrated synovial microtissues embedded in 5% Gelatin Methacrylate (GelMA). The chip integrates a pressure-driven actuator to mimic joint loading. Synovial and blood-derived RA cells were encapsulated and bioprinted, then cultured under inflammatory conditions (IL-6, TNF-α, IL-1β at 10 μg/mL). Drug testing included methotrexate and anti-TNF-α. We assessed viability, proliferation, phenotype, lubricin production, cytokine and extracellular vesicle (EV) secretion, and single-cell transcriptomics and proteomics.

**Results**

The SoC supported the formation of viable 3D microtissues replicating key RA features. Fibroblast-like synoviocytes (FLS) differentiated into lining and sublining phenotypes depending on spatial distribution. Mechanical stress upregulated lubricin secretion, while inflammation suppressed it. The platform allowed patient-specific drug testing, revealing donor-dependent cytokine modulation. Multiplex cytokine analysis and Principal Component Analysis (PCA) highlighted inter-individual variability in response to methotrexate and anti-TNF-α, supporting predictive capacity.

**Conclusion**

This autologous human SoC replicates essential aspects of RA pathophysiology and drug response. It enables rapid, personalized in vitro testing, reduces reliance on animal models, and supports the identification of patient pathotypes. Beyond therapy prediction, the SoC offers a versatile platform for studying RA heterogeneity and disease mechanisms.